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Early Development of the Australian Green Hylid Frogs

Litoria chloris, L. fallax AND *L. gracilenta*

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ABSTRACT

The life histories of *Litoria chloris, L. fallax* and *L. gracilenta* are described and shown to conform with the developmental pattern typical of Australian hylids.

INTRODUCTION

The life histories of the Australian Hylidae are poorly known. The only comprehensive life history studies of Australian hylids are those of *Litoria verreauxi* (Anstis, 1976), the *L. nannotis* species group (Liem, 1974), *L. jervisiensis* (Martin and Littlejohn, 1966), *L. burrowsi* (Martin, 1967b), *L. peroni* and *L. tyleri* (Martin *et al.,* 1979), and the *L. citropa* group (Tyler and Anstis, 1975). Tyler and Davies (1978) summarize the available information on Australian hylid life histories. Apart from the intrinsic interest of studies of life history patterns, they may also provide evidence bearing on relationships and phylogeny (Martin, 1967a, b; Martin and Watson, 1971; Watson and Martin, 1973). With this in mind we have been accumulating information on Australian hylid life histories for several years. The present account is of three species whose life histories were previously unknown: *Litoria chloris* (Boulenger), *L. fallax* (Peters), and *L. gracilenta* (Peters). The joint treatment of these three forms is not intended to imply any relationship; in their recent paper on species groupings within *Litoria,* Tyler and Davies (1978) placed *L. chloris* and *L. gracilenta* together in the *L. aruensis* group and *L. fallax* in the *L. bicolor* group.

METHODS

For *L. fallax* and *L. gracilenta,* amplectant pairs were collected in the field and placed in plastic containers with approximately 10 cm of pond water and

some grass stems for transport to the laboratory. In each case oviposition occurred in the enclosure. On return to the laboratory the embryos were reared in a constant temperature room (20 ± 0.5°C). Eggs of *L. chloris* were obtained from an amplectant pair maintained in laboratory terrarium in the Department of Zoology, University of Adelaide. Embryos were reared in a constant temperature room (27 ± 1.0°C). Samples of embryos and larvae were fixed in Tyler's fixative (Tyler, 1962) at irregular intervals. All measurements, drawings and descriptions are based on preserved material. Measurements were made using an ocular micrometer in a stereoscopic microscope, or venier calipers reading to 0.05mm. Drawings were made by accurate tracing of enlarged photographs. Other techniques used

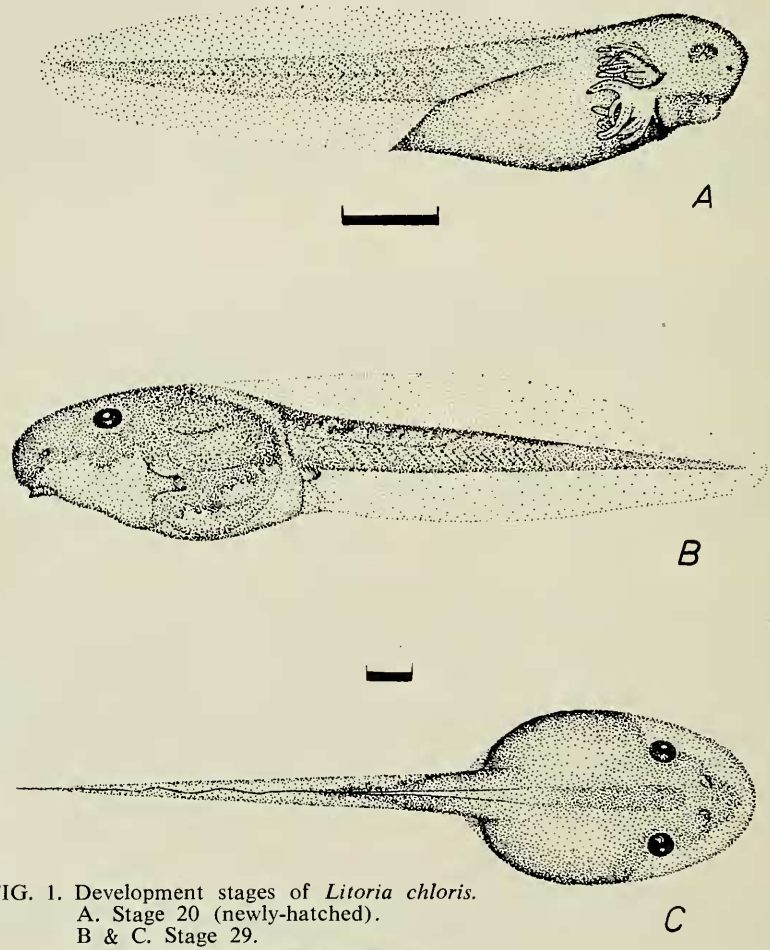

FIG. 1. Development stages of *Litoria chloris*.
A. Stage 20 (newly-hatched).
B & C. Stage 29.
The bar in each case represents 1 mm.

follow those of Martin and Littlejohn (1966), and developmental stages were classified according to the table of Gosner (1960).

RESULTS

LITORIA CHLORIS

MATERIAL:— A recently-laid clutch of eggs was obtained on 25 February, 1976, from a captive pair originally collected at Warrie N.P., Springbrook, Qld.

EGGS:— Oviposition was not observed nor could an accurate estimate of the number of eggs be obtained. Eggs were laid either singly or in very small clumps (5 - 15 eggs), loosely attached to submerged twigs. There is little colour differentiation between animal and vegetal poles with the former being mid-brown and the latter lighter brown. Each egg is surrounded by a poorly defined jelly capsule which lacks clear layers. The dimensions (mean and range) of six embryos in early cleavage (stage 9) were: embryo diameter, 1.65 mm (1.58 - 1.73); capsule diameter, 4.3 mm (3.8 - 4.7).

PRE-HATCHING EMBRYOS:— Early embryonic stages were not studied. The first series of embryos was preserved at 0830 hours on 27 February at stages 18 - 19. Their total length was about 5.3 mm. There were bulges marking the positions of the visceral arches; and optic bulges and ventral suckers were clearly visible. The overall colour was light brown with a paler yolk sac.

POST-HATCHING EMBRYOS:— Hatching began late on 27 February when the embryos were at stage 20. An embryo preserved at 0900 hours on 28 February is shown in Figure 1A. Its total length was 7.4 mm. Three pairs of external gills were present each with numerous branches. Ventral suckers were still present and the tail fin was well developed. The overall colour was mid-brown.

LARVAE:— Limb bud development began on 2 March; one larva was preserved at stage 26. The following day the larvae were at stage 29 (Figs. 1B and 1C). Pigmentation was light, the overall colour being pale brown, and the coils of the intestine were clearly visible through the body wall (Fig. 1B). The spiracle was situated below the midline on the left side and the anus opened to the right of the tail fin. The mouth disc (Fig. 2B) had a $\frac{1}{1}\ \frac{1}{2}\ \frac{1}{1}$ formula (see Martin, 1965, for explanation) and well-developed horny jaws. The papillary border was well developed and extended around the sides and back of the mouth disc. The dimensions of a stage 29 larva were: total length 15.9 mm; tail length 10.1 mm. The largest larva was at stage 39 and had a total length of 50.3 mm and a tail length of 34.7 mm.

METAMORPHOSIS:— Three individuals completed metamorphosis on 5 April. The newly-metamorphosed juveniles had body lengths of 16.3, 14.8 and 13.0 mm.

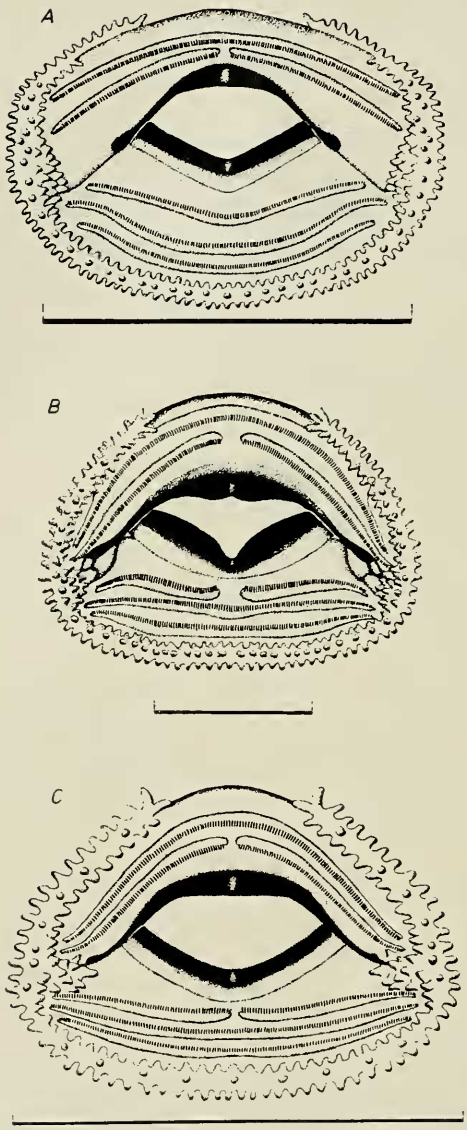

FIG. 2. Larval mouth discs at stages 27 to 29 of:
A. *Litoria fallax*,
B. *Litoria chloris*, and
C. *Litoria gracilenta*.
The bar in each case represents 1 mm.

The colour in preservative was pale brown and the general features of the juvenile resembled those of the adult (see description in Moore, 1961). All remaining specimens were preserved on 5 April.

LARVAL LIFE SPAN:— At 27°C, embryonic and larval development were completed within 41 days.

LITORIA FALLAX

MATERIAL:— A single clutch of eggs was obtained from an amplectant pair collected at Palm Grove (near Gosford), New South Wales, on 18 December 1968.

EGGS:— Oviposition was not observed. A total of 263 eggs was laid, in discrete bundles each containing 40 - 50 eggs, loosely attached to grass stems. The animal hemisphere was black and the vegetal hemisphere creamy white; each egg had a double jelly capsule. The dimensions (mean and range) of 10 embryos in early cleavage (stage 8) were: embryo diameter, 1.00 mm (0.98 - 1.03); inner capsule diameter, 1.28 mm (1.24 - 1.32); outer capsule diameter, 3.22 mm (2.80 - 3.60).

PRE-HATCHING EMBRYOS:— Early embryonic stages were not studied. The first series of embryos was preserved on 20 December, 1968, in stage 18. Their total length was about 3.2 mm. There were bulges marking the positions of the visceral arches; and optic bulges, auditory vesicles, pronephric swellings and ventral suckers were clearly visible. The overall colour was light brown, with the yolk sac creamy yellow. A further seven embryos were preserved on 23 December, when they were at stage 20 (Fig. 3A). The mean total length was 4.71 mm (range 4.44 - 5.06). Two pairs of external gills were present, the anterior pair possessing four branches and the posterior pair two. The ventral suckers were still present. The tail fin was well developed and extended along the back almost to the head. The overall colour remained light brown.

POST-HATCHING EMBRYOS:— Hatching began on 24 December, when the embryos were entering stage 21. The only marked difference from a stage 20 embryo was that the cornea was clear. The external gills were still present. On 26 December the embryos were at stage 25. The operculum was complete, with a fully developed spiracle, and the ventral suckers were still present. The mean total length was 5.43 mm (range 5.06 - 5.76).

Stage 25 was of relatively long duration, and lasted until the end of January 1969. During this stage the ventral suckers disappeared, the anal tube opened, and the mouth disc differentiated. Two stage 25 embryos preserved on 3 February 1969 have total lengths of 6.91 and 8.32 mm.

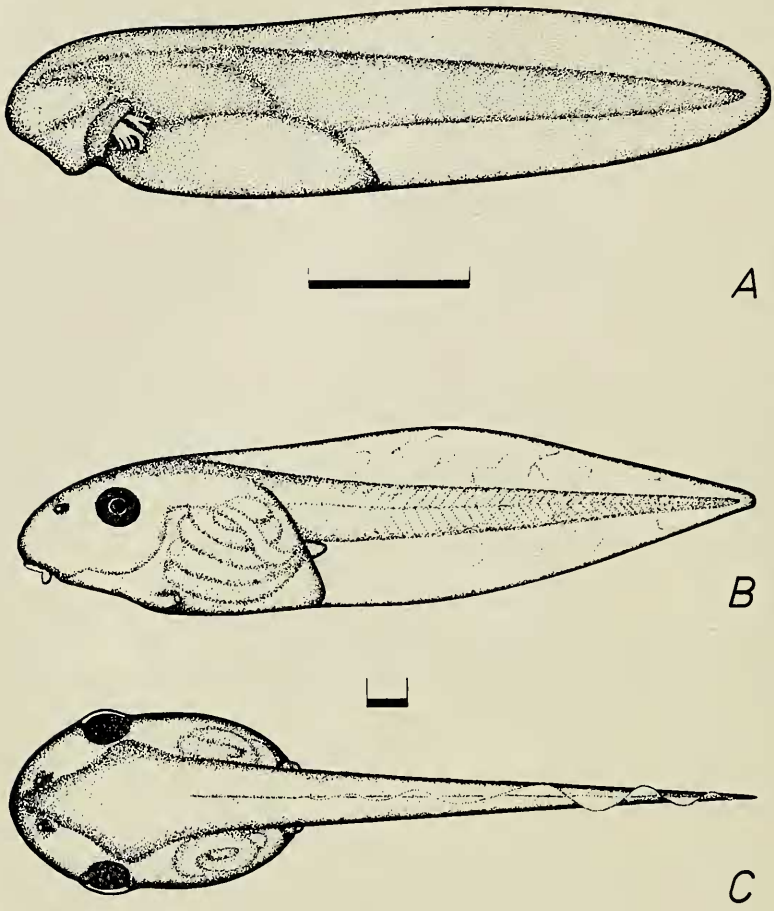

FIG. 3. Development stages of *Litoria fallax*.
A. Stage 20.
B & C. Stage 29.
The bar in each case represents 1 mm.

LARVAE:— Limb bud development was first observed on 3 February when two larvae were preserved at stage 27; their total lengths were 11.15 and 12.40 mm. On 28 February two larvae were preserved at stage 29 (Fig. 3B and C). Pigmentation was very light, the overall colour being pale yellowish brown, and the coils of the intestine were clearly visible through the body wall. The fins were clear except along some of the blood vessels. There were scattered pigment spots along the dorsal edge of the tail musculature and over the cranium. The spiracle

was situated low on the left side of the body and the anus opened to the right of the tail fin. The mouth disc (Fig. 2A) had a $\frac{1}{3}$ formula.

The papillary border extended around the sides and back of the mouth disc, and was broadened along the lateral margins. The dimensions of the two stage 29 larvae were: total length, 17.7 and 18.0 mm; tail length, 10.8 and 10.8 mm respectively.

On 27 March 1969 the larvae had reached stage 31. Two of these individuals had total lengths of 21.7 and 21.9 mm. Three larvae were preserved on 14 April when they had reached stage 41. Their body dimensions were: total length, 31.3, 32.1 and 32.4 mm; tail length, 20.5, 22.1 and 21.4 mm respectively.

METAMORPHOSIS:— Three individuals metamorphosed between 15 and 29 April 1969. These juveniles had body lengths of 10.9, 11.4 and 12.1 mm. The colouration and skin texture of the juveniles were similar to those of adults (see description in Moore, 1961).

LARVAL LIFE SPAN:— At 20°C, the larval life span extended from 118 to 132 days.

LITORIA GRACILENTA

MATERIAL:— Some eggs were obtained from a nearly-spent female collected in amplexus from a roadside pond 6.4 km SW of Nerang, Qld. on 9 November 1972.

EGGS:— Oviposition was not observed. So few eggs were laid that none were preserved. An adequate description of the egg mass was likewise impossible; the few eggs laid were loosely attached to grass stems.

PRE-HATCHING EMBRYOS:— Early embryonic stages were not studied. The first series of embryos was preserved on 11 November at stage 19. Their total length was about 3.7 mm. The tail fin was moderately well developed and bulges clearly marked the positions of the visceral arches; olfactory pits and ventral suckers were visible. The overall colour was mid-brown with a creamy yolk sac.

POST-HATCHING EMBRYOS:— Hatching began on 12 November when the embryos were at stage 20. Figure 4A shows an embryo preserved at this time. Its total length was 5.12 mm. Two pairs of external gills were present, the anterior pair having 5 - 6 branches and the posterior pair 3 - 4. The tail fin was very well developed and extended dorsally to the eye rudiment. Ventral suckers were well developed. The overall colour was pale brown with scattered dark pigment spots on the dorsum. On 13 November embryos had reached stage 24 - 25. Total lengths of two individuals preserved at this time were 6.25 and 7.19mm. At this stage the body wall was transparent revealing the coils of the yolk-filled intestine. The

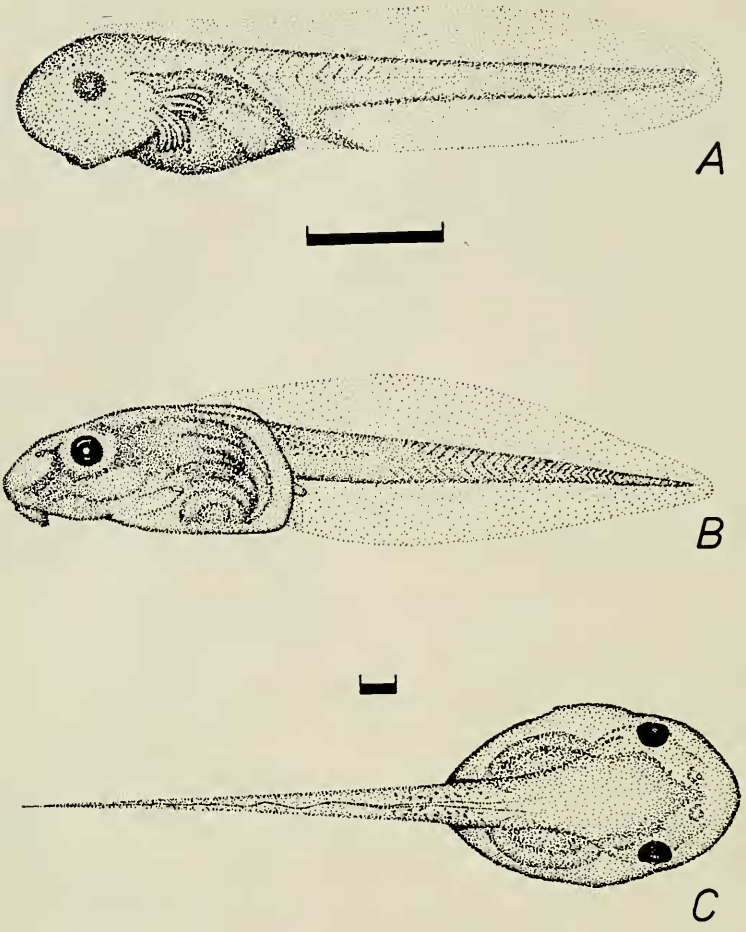

FIG. 4. Development stages of *Litoria gracilenta*.
A. Stage 20 (newly-hatched).
B & C. Stage 28.
The bar in each case represents 1 mm.

mouth had differentiated but the anus was not yet open. Ventral suckers were still present. Colour was pale brown with extensive pigment spots across the dorsum and tail musculature.

LARVAE:— The first larvae available for examination were preserved on 13 December, when they were at stage 28 (Figs. 4B and C). Dimensions of the two larvae were, total length 19.14 and 20.48 mm and tail length 11.28 and 12.00 mm

respectively. The larvae were mid-brown with dark pigmentation of the dorsum, the upper portion of the intestinal peritoneum, and the anterior tail musculature. The ventral coils of the intestine could be seen through the body wall. The spiracle was placed ventro-laterally on the left side and the anus opened to the right of the tail fin. The mouth disc (Fig. 2c) had a $\frac{1}{1}\ \frac{1}{2}\ \frac{1}{1}$ formula. The disc was bordered on its lateral and posterior margins by well developed, protruding rows of papillae.

The largest larva in this series was preserved on 9 March, 1973, at stage 37. Its dimensions were: total length 34.00mm and tail length 21.10mm. The most conspicuous difference between this individual and the stage 28 specimens was the extent of dark pigmentation. The tail fin at stage 37 had numerous large pigment spots and pigmentation was very extensive on the body and tail fin musculature.

METAMORPHOSIS:— Three individuals completed metamorphosis on 28 February 1973. These juveniles had body lengths of 11.7, 10.9 and 11.2 mm. Colouration in preservative was pale brown.

LARVAL LIFE SPAN:— At 20°C, the larval life span extended over 112 days.

DISCUSSION

As noted by Martin and Watson (1971) the life history patterns of Australian hylid frogs show remarkably little variation. The three species considered here share most of the life history characteristics typical of Australian hylids. These include aquatic oviposition, and clumped eggs, probably attached to submerged vegetation. Embryonic development and larval morphology also conform to the typical hylid pattern: well-developed external gills, a dextral anus, a sinistral, ventro-lateral spiracle, a more or less acuminate tail, and a basic 2/3 mouth formula with lateral and posterior labial papillae.

However, despite the essential conservatism of embryonic and larval characteristics in the hylids there are some noteworthy features in each of the life histories considered here. *L. chloris* is distinguished by its rate of development, which, even allowing for the difference in culture temperatures, greatly exceeds those of the other two species. This is presumably an adaptation to life in the ephemeral waters which seem to represent the typical breeding habitat of this species. Morphologically the only unusual feature of the *L. chloris* tadpole is the position of the eyes which are situated more dorsally than in the other two species, giving the tadpole a superficially leptodactylid-like appearance.

The mouth of *L. fallax* is unusual amongst Australian hylids in having three complete rows of lower labial teeth. In most Australian species the first lower row is divided.

L. gracilenta differs least from the usual hylid life history pattern, being typical of species occurring in permanent, lentic waters. It is interesting to note that while *L. chloris* and *L. gracilenta* are apparently closely related (Tyler & Davies, 1978), their patterns of larval development show adaptations to strikingly different environmental conditions.

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